

Claim Listing

1-183. (Canceled)

184. (Previously Presented) A cigarette comprising a portion of an increased-nicotine transgenic *Nicotiana tabacum* plant, wherein said cigarette is characterized by (i) a tar-to-nicotine yield ratio of between about 3 and about 8, as measured by the FTC or ISO method, and (ii) cigarette smoke having a pH of about 6.5 or lower.

185. (Previously Presented) A cigarette according to claim 184, wherein said plant expresses at least one heterologous nucleic acid that up-regulates the production of nicotine in said transgenic plant or plant portion.

186. (Previously Presented) A cigarette according to claim 185, wherein said plant expresses a heterologous nucleic acid encoding at least a segment of QPT and PMT.

187. (Withdrawn) A method of producing a nicotine buffer, comprising:

a) crossing an increased-nicotine transgenic *Nicotiana tabacum* plant with an increased-sugar *Nicotiana tabacum* plant;

b) selecting an increased-nicotine and increased-sugar *Nicotiana tabacum* progeny plant;

and

c) producing a cigarette comprising a portion of said increased-nicotine and increased-sugar *Nicotiana tabacum* plant.

188. (Withdrawn) The method according to claim 187, wherein said increased-nicotine transgenic *Nicotiana tabacum* plant is produced by:

a) transforming a *Nicotiana tabacum* plant with a construct comprising, in the 5' to 3' direction, a promoter operably linked to a heterologous nucleic acid encoding an enzyme or transcription factor that up-regulates nicotine synthesis;

b) regenerating transgenic *Nicotiana tabacum* plants from the transformed plant; and

c) selecting a transgenic *Nicotiana tabacum* plant having increased-nicotine content relative to a non-transformed control plant.

189. (Withdrawn) A method for producing a nicotine buffer, comprising

a) providing a transgenic *Nicotiana tabacum* plant that expresses a heterologous nucleic acid sequence that confers elevated nicotine and fatty acid levels, relative to a non-transformed control plant, and

b) producing a cigarette that comprises a portion of said plant.

190. (Withdrawn) The method according to claim 189, wherein step (a) comprises:

1) transforming a *Nicotiana tabacum* plant with (i) a first construct comprising, in the 5' to 3' direction, a promoter operably linked to a heterologous nucleic acid encoding an enzyme or transcription factor that up-regulates nicotine synthesis; and (ii) a second construct comprising, in the 5' to 3' direction, a promoter operably linked to a heterologous nucleic acid encoding an enzyme or transcription factor that up-regulates fatty acid synthesis

2) regenerating transgenic *Nicotiana tabacum* plants from the transformed plant; and

3) selecting a transgenic *Nicotiana tabacum* plant having increased-nicotine content and increased fatty acids relative to a non-transformed control plant.

191. (Withdrawn) A method according to claim 189, wherein step (a) comprises:

a) crossing an increased-nicotine transgenic *Nicotiana tabacum* plant with an increased fatty acid *Nicotiana* transgenic plant;

b) selecting an increased-nicotine and increased fatty acid *Nicotiana tabacum* progeny plant;

and

c) producing a cigarette comprising a portion of said progeny plant.

192. (Withdrawn) A method for increasing nicotine and cured reducing sugar content in a *Nicotiana tabacum* plant, comprising:

- a) transforming a *Nicotiana* plant having elevated cured reducing sugar content with a nucleic acid conferring an increased-nicotine phenotype;
- b) regenerating transgenic *Nicotiana tabacum* plants from the transformed plant; and
- c) selecting a progeny plant having increased cured reducing sugar content and increased-nicotine content relative to a non-transformed control plant.

193. (Withdrawn) A method of increasing alkaloid biosynthesis in a *Nicotiana tabacum* plant or part thereof, comprising:

- a) transforming a *Nicotiana tabacum* plant with at least one nucleic acid sequence, wherein said nucleic acid increases quinolate phosphoribosyl transferase and putrescine N-methyltransferase expression;
- b) regenerating transgenic *Nicotiana tabacum* plants from the transformed plant; and
- c) selecting a plant having increased alkaloid biosynthesis, wherein said plant has increased quinolate phosphoribosyl transferase and putrescine N-methyltransferase expression relative to a non-transformed control plant.

194. (Withdrawn) A tobacco plant characterized by increased nicotine levels and high cured reducing sugar content, wherein at least one transgene confers the increased nicotine levels.

195. (Withdrawn) A tobacco plant according to claim 194, wherein said plant has a nicotine content greater than 3.5 percent and a cured reducing sugar content from between about 12 percent and about 30 percent.

196. (Withdrawn) A method of making a tobacco product having lower levels of tobacco-specific nitrosamines, comprising:

a) providing a reduced nicotine transgenic *Nicotiana tabacum* plant or plant portion, wherein said plant or plant portion expresses at least one heterologous nucleic acid that down-regulates nicotine production;

b) producing tobacco from said transgenic plant or plant portion;

c) adding nicotine to said tobacco; and

d) producing a product comprising said tobacco, wherein said product has a lower level of tobacco-specific nitrosamines relative to a control product comprising a non-transformed control plant or plant portion.

197. (Withdrawn) A product produced by the method of claim 196, wherein said product is cigarette tobacco.

198. (Withdrawn) The tobacco product of claim 197, wherein said cigarette tobacco comprises filler having a level of tobacco-specific nitrosamines below about 0.5 microgram per gram of tobacco.

199. (Withdrawn) The tobacco product of claim 198, wherein said level of tobacco-specific nitrosamines is less than 0.05 microgram (50 ppb) per gram of tobacco.

200. (Withdrawn) A method of making reconstituted tobacco, comprising:

a) providing plant material selected from a group consisting of

(i) an increased-nicotine transgenic *Nicotiana tabacum* plant or plant portion, wherein said plant or plant portion expresses at least one heterologous nucleic acid that up-regulates the production of nicotine in said transgenic plant or plant portion;

(ii) a reduced-nicotine transgenic *Nicotiana tabacum* plant or plant portion, wherein said plant or plant portion expresses at least one heterologous nucleic acid that down-regulates the production of nicotine in said transgenic plant or plant portion;

(iii) deproteinized tobacco fiber or tobacco fiber portion; and

(iv) freeze-dried tobacco or tobacco portion; and

b) combining and reconstituting any combination of said plant material.

201. (Withdrawn) A cigarette comprising the reconstituted tobacco of claim 200, wherein said cigarette has a reduced yield of compounds selected from a group consisting of polycyclic aromatic hydrocarbons, tobacco-specific nitrosamines, benzo(a)pyrene, phenols, and catechols relative to a cigarette made from a control plant.